ATTORNEY'S DOCKET CS-120
PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re the application of:	)	Group Art Unit: 1644
	)	
TALOR	)	Examiner: Unknown
	)	
Serial No. 10/611,914	)	
	)	
Filed: July 03, 2003	)	

For: A METHOD OF PRE-SENSITIZING CANCER PRIOR TO TREATMENT WITH RADIATION AND/OR CHEMOTHERAPY AND A NOVEL CYTOKINE MIXTURE

DECLARATION UNDER 37 CFR 1.132 OF EYAL TALOR

Commissioner for Patents
Alexandria, VA 22313-1450

Sir:

I, Eyal Talor, do hereby declare and state as follows:

1. I am a citizen of the United States of America.

2. I am the inventor of the invention disclosed and claimed in the above-identified application.

3. In 1987, I received a Ph.D. in Microbiology and Immunology from the University of Ottawa (Ontario, Canada) and completed a Post-Doctoral Fellowship at Johns Hopkins University in 1998 and have been involved in cellular immunity, autoimmunity, infectious disease and cancer research since the mid 1980's.

4. My Curriculum Vitae is attached hereto.

5. In order to demonstrate that the novel cytokine mixture is a unique mixture of cytokines having only trace amounts of IL-12 and an IL-8 to IL-2 ratio of 411, the following

experiments were conducted and carried out by me or under my directions and supervision.

Experimentation

The following experiment provides a characterization chart of a novel mixture of cytokines by ELISA (Enzyme Linked Immunosorbant Assay) of both detected and undetected components in the cytokine mixture. The experiment ascertains the characteristics of different lots of the novel cytokine mixture as measured by commercially available cytokine assays (ELISA assays).

In particular, the cytokine mixture characterization Table 1 shows that the mean value of IL-8 in the cytokine mixture is 164,404 pg/mL. The mean value of IL-8 (164,404) divided by the label claim for IL-2 in the finished product, 400 IU of IL-2 (where IU represents International Units for Interleukin-2 given in World Health Organization 1st International Standard for Human IL-2, 86/504) results in an IL-8 to IL-2 mean ratio of 411.

Table 1 also shows that the cytokine mixture only has trace amounts of IL-12 having a mean value of 42 pg/mL. The range of IL-12 in the cytokine mixture is 28 pg/mL to 67 pg/mL with the

mean value of 42 pg/mL where the number of lots of the finished product sample is five.

1. Material and Methods

1.1 Cytokine mixture

The novel cytokine mixture was produced in accordance with Master Batch Records developed specifically for the manufacture of the novel cytokine mixture under Current Good Manufacturing Practice (cGMP) provided in 21 C.F.R. § 211.186 for the manufacture of a product for human use.

Three independently manufactured lots of the cytokine mixture were produced and tested independently for the presence or absence of each of the different commercially available cytokines and small biological molecules.

1.2 Cytokine and other small biological molecules ELISAs used in the characterization of the novel cytokine mixture

Commercially available cytokines and other small biological molecules ELISA assays were purchased from commercial vendors and assessed in house for their ability to measure a known amount of analyte such as cytokine or small biological molecules (thought to be present in the novel cytokine mixture). These included the

following ELISA assays: IL-1α, IL-1β, IL-2, IL-3, IL-4, IL-6, IL-7, IL-8, IL-10, IL-12, IL-15, IL-16, TNF-α, IFN-γ, GM-CSF, TNF-β, SCF, LIF, 6k-PGF1α, Angiogenin, FGF-basic, sE-Selectin, EGF, EPO, G-CSF, sICAM-1, IFN-α, LTB4, LTC4, MIP-1α, MIP1-β, PDGF-AB, PGE_2, TfR, TGF-β_2, TxB2, and RANTES.

1.3 ELISA Assay Testing Procedure

In order to ascertain the presence or absence of a particular cytokine or small biological molecule in the novel cytokine mixture, each lot of the finished product as well as the manufacturing intermediates for the same lot were tested in the cytokine and other small biological molecule ELISA assays. Each lot of the three independently produced lots of the cytokine mixture was tested at least twice in at least two independent ELISA assays. Each ELISA assay was performed independently of the other ELISA assay. The analysis of each ELISA assay's quantitative results is set forth in Tables 1-3.

2. Results

2.1 Table 1

The following detected cytokines and small biological molecules in the cytokine mixture are shown as follows in the

Table 1.

Table 1

Detected in the cytokine mixture	Mean pg/mL[(a)]
IL-1α	300
IL-1β	443
IL-2	405[(b)]
IL-3	211
IL-6	18,208
IL-8	164,404
IL-10	1,219
IL-12	42
IL-16	735
TNF-α	2,913
IFN-γ	2,082
GM-CSF	1,389
TNF-β	723
6k-PGF1α	61
EGF	109
G-CSF	1,188
sICAM-1	3.5[(c)]
LTB4	15
MIP-1 α	12,347
MIP-1 β	9,103
PDGF-AB	60
PGE_2	1,819
TfR	3.5 [(c)]
TxB2	9,718
RANTES	1,008

(a) Measured by Commercially available ELISA
(b) Expressed as IU/mL (Measured, Label claim is 400 IU/mL)
(c) Expressed in ng/mL

2.2 Table 2

The following undetected or below detection levels of cytokines and other small biological molecules in the cytokine mixture are shown in the Table 2.

Table 2

Not detected in the cytokine mixture	Mean pg/mL[(a)]
IL-4	<31[(b)]
IL-7	<16[(b)]
IL-15	<3.9[(b)]
SCF	<31[(b)]
LIF	<31[(b)]
Angiogenin	<78[(b)]
FGF-basic	<10[(b)]
sE-Selectin	<600[(b)]
EPO	<20 [(b)] [(c)]
IFN- α	<26[(b)]
LTC4	<25[(b)]
TGF- β_2	<31[(b)]

(a) Measured by Commercially available ELISA
(b) The limit of detection of the specific ELISA assay
(c) Expressed in mIU/mL

2.3 Table 3

The ratio range of various cytokines and small biological molecules to IL-2 are shown in Table 3 where IL-2 was fixed at the label claim of 400 IU/mL, IU being International Units for Interleukin-2 given in World Health Organization 1st

International Standard for Human IL-2, 86/504.

Table 3

Cytokine present in the cytokine mixture	Ratio range to IL-2
IL-1α	0.56 - 0.94
IL-1β	0.4 - 1.5
IL-2	1.0 (by definition)
IL-3	0.38 - 0.68
IL-6	37.2 - 53.8
IL-8	261 - 561.5
IL-10	2.82 - 3.22
IL-12	Trace[(a)]
IL-16	1.16 - 2.84
TNF-α	3.2 - 11.3
IFN-γ	1.5 - 10.9
GM-CSF	2.2 - 4.8
TNF-β	1.17 - 2.43
6k-PGF1α	Trace[(a)]
EGF	0.267 - 0.283
G-CSF	2.16 - 3.78
sICAM-1	Trace[(a)]
LTB4	Trace[(a)]
MIP-1 α	15.7 - 37.16
MIP-1 β	17.1 - 28.5
PDGF-AB	Trace[(a)]
PGE_2	3.63 - 5.42
TfR	Trace[(a)]
TxB2	23.47 - 25.13
RANTES	2.3 - 2.7

(a) Trace = Detected at levels just above the level of detection of the specific ELISA assay (in the matrix, "The Novel Cytokine Mixture", Leukocyte Interleukin Injection - finished product).

CONCLUSION

Based on the experiments above, it has been demonstrated that the mean value of IL-8 (164,404) divided by 400 IU of IL-2 is 411.

It has also been demonstrated that the cytokine mixture only has trace amounts of IL-12 having a mean value of 42 pg/mL wherein the value of 42 pg/mL is in the range of 28-67 pg/mL.

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Eyal Talor, Ph.D.

Feb 18, 2004.
DATE

CURRICULUM VITAE

EYAL TALOR, Ph.D.
CEL-SCI Corporation
Phone (work): 410-358-6866
Fax (work): 410-358-1647
e-mail: etalor@cel-sci.com

Dr. Talor is a clinical cellular immunologist with over 18 years of clinical research and development and industrial manufacturing experience. He has significant expertise in managing clinically oriented research and development of drugs for immunotherapy application. His expertise includes cGMP manufacture, Quality Control testing, and the design and build out of GMP manufacturing and testing facilities. He also served as Director of Clinical Laboratories (certified by the State of Maryland) and has experience in the design of clinical trials and GCP requirements. Dr. Talor has broad experience in the different aspects of product assay and development, analytical methods validation, raw material specifications, and QC tests development under FDA/GMP, U.S.P., and ICH guidelines. He has extensive experience in the preparation of documentation for IND and other regulatory submission. His areas of expertise encompass immune response assessment, and he has published a number of reviews on immune regulations in relation to clinical immunology.

EDUCATION

Post Doctoral Fellow, Department of Immunology and Infectious Diseases, the Johns Hopkins University, School of Hygiene and Public Health, **1987-1988**
Ph.D. (Summa Cum Laude), Immunology and Microbiology, University of Ottawa, **1987**
B.S. (Cum Laude with honors), Animal Physiology, University of Ottawa, **1983**
B.S., Biology, University of Ottawa, **1982**

PROFESSIONAL HISTORY

CEL-SCI Corporation	**-Senior Vice President** of Research and Manufacturing **1998- Present** **-Vice President** of Research and Manufacturing **1994 - 1998** **Scientific Director** and Secretary PRAL Laboratories **1994 - 1997**
CBL Inc., (CEL-SCI Corp.)	**-Director**, Research Manufacturing and Quality Control **1993 - 1994** **-Director**, Clinical Laboratory **1993 - 1994**
SRA Technologies, Inc.	**-Principal Scientist**, **1991 - 1993** **-Director**, Flow Cytometry Laboratory, **1991 - 1993** **-Director**, Clinical laboratory (Certified, State of Maryland, USA), **1992 - 1993** -Regulatory Affairs and Safety Officer, **1992 - 1993**
The Johns Hopkins University	**Academic Appointments, 1987 - Present** Department of Molecular Microbiology and Immunology -Research Associate and Lecturer, **1987 - 1991** -Adjunct Associate, **1991 - Present**
Ottawa Civic Hospital	-Teaching Assistant, **1985 - 1986**
University of Ottawa	-Teaching Assistant, **1983 - 1984**
Center for Technical Education (under the auspices of the Tel-Aviv University)	-Research Coordinator, **1978**

EXPERIENCE

As Senior Vice President of Research and Manufacturing for CEL-SCI Corporation, Dr. Talor is charged with all research and manufacturing activities and scientific operations of the company, including the overseeing of the R&D, manufacturing and scale up, under cGMP, of parenterals for clinical investigational use. Dr. Talor is also in charge of overseeing QC, process and assay development, validation, and drug product and biologics, and tissue culture research and development. Dr. Talor has made a number of discoveries and proprietary (trade secrete) improvements to both product and process, including specialized process and fill/finish equipment. He designed a bulk manufacturing and a fill and finish facility for the product. He also designed, oversaw the construction, build out and equipping of PRAL laboratories, CEL-SCI corporation's QC, R&D, and service laboratory. As Scientific Director, he was in charge of all the scientific and administrative operations of the laboratory. At PRAL laboratories, Dr. Talor was also in charge of assay development, validation, and testing services for industry, government and academia under GMP/GLP. Dr. Talor has managed multi-million-dollar budgets and up to 40 professionals.

As the senior scientific officer of the Company, Dr. Talor is involved in business development, and is a point of contact for negotiations of product and process assessment and product/process acquisition and in-licensing activities.

In the capacity of Director of Research, Manufacturing and Quality Control and Director of the Clinical Laboratory, at CBL Inc., (in support of CEL-SCI Corp.) Dr. Talor directed the scientific and administrative operations of a manufacturing pilot facility involved in the manufacture of parenterals for clinical investigation. He also headed the research and development of various scientific methods and assays in support of product QC. In addition, he headed the process development team in support of product development.

As a Principal Scientist with SRA technologies, Inc., Dr. Talor directed the scientific and administrative operations of a multi-million dollar contract with the U.S. armed forces, managing a team of 23 research professionals. In this capacity he lent his expertise in immunolobiology for the development and assessment of immunocompetence in retroviraly infected patients. He developed and utilized cellular and humoral immune tests for the evaluation of patients undergoing therapy for acquired immunodeficiency disease syndrome.

While at SRA, Dr. Talor design, implemented, and directed a BL-3 flow cytometry facility with efforts concentrated in cell phenotyping and cell sorting of HIV infected patients. In addition, he functioned as Director of Clinical Laboratory and was involved with CAP, FDA, and CDC accreditation and monitoring programs. In the capacity of Regulatory Affairs and Safety Officer, Dr. Talor was in charge of standards, certification, development and implementation of quality control and quality assurance programs and compliance with CAP, FDA, CDC, State, and GLP requirements.

Before coming to SRA, Dr. Talor directed the technical and administrative operation of the Immunology and Infectious Diseases laboratory at the Johns Hopkins School of Hygiene and Public Health. He directly supervised the senior technical staff and the postdoctoral fellows in the laboratory. As an expert in cellular immunology and particularly immunoregulation, he was regularly consulted by other investigators both from within the Johns Hopkins School of Public Health and School of Medicine and outside of the University on the many aspects of immune regulation.

While at Johns Hopkins School of Public Health, Dr. Talor developed an *in vitro* assay to assess immune suppression *in vivo* with the use of cell transfer to SCID mice. He also was involved in research and development of a field assay kit for the detection of schistosomal secerial antigen in Third World Water reservoirs. Other research interests of his are the effect of UVB exposure *in vivo* on the immune response in humans. In this instance he developed an *in vitro* assay that was correlated to the *in vivo*

effect of UVB on the immune system of humans. Through collaborative work, in a team lead by Dr. Talor, he was involved in the development of a biologically derived suppressor factor for use in humans (at University of Ottawa.)

Dr. Talor is committed to the enrichment of educational programs in immunology and is involved in the teaching of immunological aspects in various courses in the Department of Molecular Microbiology and Immunology at the Johns Hopkins Bloomberg School of Public Health. He also coordinates and teaches a course in clinical immunology at the graduate level at the Johns Hopkins School of Public Health in conjunction with the School of Medicine.

Prior to his academic career, Dr. Talor was a Research Coordinator at the Center for Technical Education (under the auspices of the Tel-Aviv University). In that capacity he coordinated the testing and evaluation of scholastic aptitude tests, administered to high school students, managing 30 professional personnel.

ADDITIONAL TRAINING

Paramedical Training, Israeli Defense Forces (IDF), Israel, **1978**
Psychometric Tester, Chief Coordinator of the Evaluation of Air Force Pilot Candidates, IDF, Israel, **1975-1978**
General Survey of Physics and Computers, Summer Course, Bar-Ilan University, Israel, **1973**
Youth Scientist Course, Microbiology, Bar-Ilan University, Israel, **1972-1973**

PROFESSIONAL AFFILIATIONS

American Association of Immunologists
Clinical Immunology Society
American Association for the Advancement of Science

PATENTS and INVENTIONS

Patents:
US Patent Application No. 10/111,602: "Peptide Constructs for Treating Autoimmune and Related Diseases"

US Patent Application No. 10/682,979:"A Standardized Serum Free and Mitogen Free Cytokine Mixture and a Production Process Thereof"

US Patent Application No. 10/611,914: "A Method of Pre-Sensitizing Cancer prior to Treatment with Radiation and/or Chemotherapy and a Novel Cytokine Mixture"

US Provisional Patent Application: "Method of Treating Diseases of Viral Ethiology with Immuno-Boosting Compounds"

Inventions: Numerous (Trade Secrets) process and product

OTHER ACTIVITIES

- **Member**, Board of Directors, BZD, Baltimore, **1998-Present**
- **Member**, Board of Directors, PRAL, Laboratories, Inc., **1994-1997**
- **Executive Board Member**, Board of Directors, U.S. -Israeli Biotechnology Council, **1995-2001**
- **Executive Board Member**, Greater Baltimore Committee, Technology Council, **1995-1997**
- **Member**, Greater Baltimore Committee, Technology Transfer Committee, **1995-1997**
- ***Ad hoc* Reviewer**, Cellular Immunology, **1987-Present**
  The Journal of Immunology, **1987-Present**
  Clinical Immunology and Immunopathology, **1988-Present**
  European Journal Of Immunology, **1990-Present**
  Vaccine, **2001-Present**

SELECTED PUBLICATIONS

1) **Talor, E**. and Rose, N.R.: The aging thymus: Loss of specific regulation increases autoimmunity (in preparation).

2) **Talor, E.**, Rose, N.R., Sharma, R., Strickland, P.T., Lesko, S.A., Jr., and Ts's, POP: UVB exposure *in vitro* of human peripheral blood mononuclear cells causes immune parameter changes which correlate with immune response changes after UVB exposure *in vivo* (in preparation).

3) Timar, J., Forster-Horvath Cs., Lukits J., Dome B., Ladanyi A., Remenar E., Kasler M., Bencsik M., Repassy G., Szabo Gy., Velich N., Suba Zs., Elo J., Balatoni Zs., Bjtai A., Falus A., Pallinger E., Chretien P., and **Talor E.** The Effect of Leukocyte Interleukin, Injection on Peri- and Intratumoral Subpopulation of Mononuclear Cells and on Tumor epithelia - An Alternative Approach to Assessing Response to Treatment - a Multi-Center Phase II Clinical Trial (in preparation).

4) Timar, J., Forster-Horvath Cs., Lukits J., Dome B., Ladanyi A., Remenar E., Kasler M., Bencsik M., Repassy G., Szabo Gy., Velich N., Suba Zs., Elo J., Balatoni Zs., Bjtai A., Chretien P., and **Talor E.** The Effect of Leukocyte Interleukin, Injection (Multikine®) Treatment on the Peritumoral and Intratumoral Subpopulation of Mononuclear Cells and on Tumor epithelia: A possible New Approach to Augmenting Sensitivity to Radiation and Chemotherapy in Oral Cancer - A Multi Center Phase I/II Clinical Trial. The Laryngoscope 113:2206, December 2003.

5) Zimmerman, D.H., Lloyd, J.P., Heisey, D., Winship, D.H., Siwek, M., **Talor, E.,** and Sarin, P.S: Induction of Cross Clade Reactive Antibodies in Mice by Conjugates of HGP-30 (Peptide Analog of HIV-1sf2 p17) and Peptide Segments of Human β-2-Microglobulin or MHC II β chain. Vaccine 19: 4750-4759, 2001.

6) Taylor, G., Ely, L., Wolff, T., Davis, C., Ioffe, O., **Talor, E**., Redfield, R., and Tramont, E: Immunotherapy with Leukocyte Interleukin, Injection for Human Papilloma Virus (HPV) Induced Cervical Dyspalsia in HIV Patients. October 2001, Ann. Of the International Society for Interferon and Cytokine Research.

7) Brown, S., Fareed, G., **Talor, E.,** and Winship D: A Phase I Open-Label Study of Leukocyte Interleukin, Injection in HIV-1 infected individuals: Preliminary Evidence for Improved Delayed Type Hypersensitivity Responses to Recall Antigens. Antiviral Therapy 5(supplement) 18, 2000.

8) Harris, D.T., Matyas, G.R., Gomella, L.G., **Talor, E.,** Winship, M.D., Spitler, L.E., and Mastrangelo, M.J: Immunologic Approaches to the Treatment of Prostate Cancer, Seminars in Oncology Vol 26, No 4 (August) 439-447, 1999.

9) Sarin P.S., Lloyd J., Heisey, D., **Talor, E**., Winship, D., Zimmerman, D.: HGP-30W vaccine induces cross clade (A,B,C,D,&E) recognition and amplification of cellular immune responses (TH-1) with different adjuvants and in a prim-boost protocol. Proceedings of the 12th World AIDS Conference, Switzerland, 1998.

10) Cherigos, M.A., **Talor, E**., Sidwell, R. W., Burger, R.A., and Warren, R.P.: Leukocyte Interleukin, Injection (LI) augmentation of natural killer cells and cytolytic T-lymphocytes, Immunopharmacology and Immunotoxicology 17(2): 247, 1995.

11) Richter, M., Kaplan, H., Kragg, G., **Talor, E.**, and Jodouin, C.A.: A non-cytrotoxic suppressor of immunoglobulin synthesis and secretion by "B" cells of normal humans and patients with rheumatoid arthritis and systemic lupus erythematosus. Autoimmunity 11:107, 1991.

12) Rose, N.R. and **Talor, E.**: Antigen-specific Immunoregulation and Autoimmune Thyroiditis. Ann. NYAS 636:306, 1991.

13) Rose, N.R. and **Talor, E.**: Antigen-specific thymus-derived suppressor cells from mice injected with mouse thyroglobulin. Immunol. 1991.

14) **Talor, E.** and Rose, N.R.: A Hypothesis: The aging paradox and autoimmune disease. Autoimmunity. 8:245, 1991.

15) Richter, M., Trudel, I., and **Talor, E.**: Cell involved in the immune response XXVIII. Suppressor cells in the spleen of the outbred rabbit following primary immunization are T cells which migrate from the thymus. Scan. J. Immunol. 36-611, 1990.

16) **Talor, E.** and Rose, N.R.: Suppressor cells and suppressor factors in the regulation of humoral immunity. In: Contemporary Clinical Immunology, ed. By M.R. Escobar. Plenum Press, NY, 1989.

17) **Talor, E.** and Rose, N.R.: Antigen-specific thymus-derived suppressor cells from mice injected with mouse thyroglobulin. (J.G. Kaplan, D.R. Green, and R.C Bleackley eds.) Alan R. Liss, Inc., NY, pg. 391, 1989.

18) **Talor, E.** and Rose, N.R.: The suppression of splenic antibody-secreting cells by mouse thyroglobulin-primed thymocytes *in vitro*. Clin. Immunol. Newsletter, 1989.

19) Newmann, D.A., Lane, J., **Talor, E.**, Herskowitz, A., and Rose, N.R.: Antibody and antibody secreting cells in the hearts of mice with experimental autoimmune myocarditis. Proceedings of the International Symposium on Inflammatory Heart Disease, 1988.

20) **Talor, E.**, Joduin, C.A., and Richter M.: Cells involved in the immune response XXXVI. The thymic antigen-specific suppressor cell in the immunized rabbit is a T cell with receptors for F G and the antigen and it acts, via a secreted suppressor factor, directly on the immune splenic AFC B cell to inhibit antibody secretion. Immunol. 64:253, 1988.

21) **Talor, E.** and Richter M.: Cells involved in the immune response XXXV. The antigen-specific antibody response in the rabbit is suppressed by thymocytes of allogeneic immunized rabbits and by the non-toxic suppressor factor (NTSF) secreted by these thymocytes. Clin. Immunol. Immunopath. 49(2):150, 1988.

22) **Talor, E.** and Richter M.: Suppressor factors of non-T Cell origin in the regulation of humoral immune response in man. Clin. Immunol. Newsletter. Vol-9 Nov. 1, 1988.

23) **Talor, E.** and Richter M.: T cell derived suppressor factors in the regulation of humoral immunity. Clin. Immunol, Newsletter. Vol 9. No. 4, 1988.

24) **Talor, E.** and Rose N.R.: Antigen-specific mouse thyroglobulin-primed thymocytes suppress syngeneic splenic antibody secreting cells *in vitro*. Fed. Proc., 1988.

25) **Talor, E.** and Rose N.R.: En route to the regulation of autoimmune thyroid disease: Autoantigen-specific suppression. Proceedings of the International Symposium on Immunointervention, Paris, 1988.

26) **Talor, E**. and Rose N.R.: The induction of antigen-specific thymic regulatory cells in the mouse. Cellul. Immunol. 116:24, 1988.

27) **Talor, E**. and Richter M.: Suppressor cells in the regulation of humoral immunity in humans. Clinical. Immunol. Newsletter. Vol.8. No. 11, 1987 (Also translated and appears in the Japanese version of Clinical Immunology Newsletter, Japan).

28) Richter, M. And **Talor, E**.: Cells involved in the immune response XXXIV. Suppressor cells in the thymus of the rabbit capable of secreting a factor which can suppress the secretion of antibodies from antibody-forming cells *in-vitro*. Clinical Immunol. and Immunopath. 41:461, 1986